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UNPUBLISHED PRELIMINARY DATA

Final Report

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HISTOLOGICAL STUDIES ON THE INNER EARS OF SELECTED MONKEYS

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I. Introduction

The purpose of this grant has been the histological preparation of selected squirrel monkey ears. The funds were used entirely for the purchase of supplies and technicians' salaries in the histological laboratory in the department of Otolaryngology, Washington University Medical School, under the supervision of Dr. Walter P. Covell.

The monkeys had been previously treated with streptomycin sulfate by Dr. Ashton Graybiel, MC, USN, at the U.S. Naval School of Aviation Medicine in Pensacola, Florida. At appropriate intervals, they were tested for canal sickness, and given caloric tests there. They were then shipped to St. Louis for fixation of the temporal bones.

II. Preparation Procedure

a. Fixation

The monkey ears were fixed by means of intra-arterial perfusion. The monkeys were anaesthetized by intra-peritoneal injection of a slight excess of nembutal (0.2 cc-0.4cc). The pleural cavity was then opened and a cannula inserted into the left ventricle for perfusion. The right auricle was snipped to allow outflow of blood and fluid. The blood was first washed out with a small amount of 0.9% sodium chloride. This was immediately followed with Heidenhain-Susa fixative, prepared according to the formula given by Eggston and Wolff. (Eggston, A.A. and Wolff, D.: Histopathology of the Ear, Nose, and Throat. Williams and Wilkins Co., Baltimore, 1947.) The temporal bones were then removed and immersed in the fixative for 18 to 24 hours.

b. Decalcification and Embedding

1- 95% alcohol for 12 hours.

2- Decalcify in 3% HCL, with solution changes each day. Decalcification was complete in approximately 5 days.

3- Wash in running tap water, 18 to 24 hours.

4- Neutralize in 5% Na_2SO_4, 12 to 24 hours.

5- Dehydrate in 80%, 95%, 100% ethanol and equal parts ether-alcohol, 12 to 18 hours each.

6- Embed in celloidin, 2%, 4%, 8% and 12%, one week each.

The celloidin solvent (ether-alcohol) was then allowed to evaporate slowly for the final formation of the hardened block of celloidin with the temporal bone, properly oriented, embedded in it.

The sections were cut on a Spencer sliding microtome at approximately 15 microns thickness. Serial sections were made of each temporal bone in entirety. Every fifth section was stained routinely either with hematoxylin and eosin, or Mallory's triple connective tissue stain.

III. Material

Eleven squirrel monkeys were received from Pensacola. Three were normal controls; eight were streptomycin treated. One of the normals expired before it could be perfused. This was replaced with a normal squirrel monkey whose eyes had been removed for other purposes. The table shows the number of sections made for each ear and the stain used. Monkeys OPH, OK and OG were the normal controls.

Monkey	Number of sections cut		Number of sections stained		Stain	
	Right	Left	Right	Left	Right	Left
OPH	175	250	36	50	H&E	H&E
OK	200	240	40	36	H&E	H&E
BV	200	200	40	40	H&E	H&E
BT	240	270	48	54	H&E	H&E
EH	200	200	40	40	H&E	H&E
BM	475	360	95 12	75	M H&E	H&E
AL	400	320	80	64	H&E	H&E
AC	360	360	47 25	16 52	M H&E	M H&E
AG	210	120	18 42	33 42	M H&E	M H&E
AA	200	180	40	36	H&E	H&E
OG	200	220	40	44	H&E	H&E

M: Mallory triple connective tissue stain.

H&E: Hematoxylin and eosin stain.

IV. Findings

The sections from the ears of all the animals in the table have been examined. Studies have been made on the sensory areas in the vestibule, and presence and condition of the hair cells noted. Alterations in the organ of Corti have also been studied. Further observations are being made under N.I.H. support, and the complete analysis will be published in one of the professional journals with due credit to both NASA and NIH. A summary is given below:

1. Vestibule:

a- Cristae of the semicircular canals.

The animals may be divided into two groups with respect to sensory cell damage, ie: those which show marked loss of hair cells, and those showing pathology but little or no hair cell loss.

Animals EH, BT and BV belong to the first group. The hair cell loss in these animals was estimated to range from 50% to 80%. It appears to begin at the top of the cristae and then progress along the sides toward the planum semilunatum. The supporting cells appear lower in height, but whether this actually represents a pathological change, or merely realignment of the cell, cannot be determined.

Animals AA, AC, AL, AG, and BM belong to the second group. The hair cells show nuclear pyknosis and cytoplasmic shrinkage. Around the altered hair cell is a wide pale halo, and this is believed to be the nerve chalice, swollen to about twice its normal size.

b- Otolithic Organs

The maculae of utricle and saccule show very little pathological change, and in each ear it was always considerably less than the pathology in the cristae. In some animals, as for example BT, both utricle and saccule appeared essentially normal. In other animals, such as AA, intermittent nuclear pyknosis and hair cell shrinkage with accompanying nerve chalice

swelling was present.

2. Organ of Corti:

All of the animals, except AA, showed some loss of external hair cells from the organ of Corti. This was always limited to the first turn. Many of the ears showed a complete loss of the organ of Corti at its vestibular end near the round window. Hair cell distortion or swelling continued up into the second turn in some ears, but in no case was any injury visible in the upper turns of the cochlea.

V. Summary

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The ears from eight squirrel monkeys treated with streptomycin sulfate, and three normal monkeys were perfused, fixed, embedded in celloiden, cut and stained. All the streptomycin treated animals showed either pathology or hair cell loss in the cristae of the semi-circular canals. The otolithic organs were either normal in appearance or showed minor changes. The organ of Corti of each, except AA, showed changes ranging from outer hair cell loss to complete degeneration, limited primarily to the first turn of the cochlea.

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